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AMENDMENTS TO THE SPECIFICATION:

Pursuant to the proposed revisions to 37 C.F.R. § 1.121, please amend the specification as follows:

Please replace the paragraph beginning at page 6, line 14, with the following two rewritten paragraphs:

B1
-- Figure 7 shows that the chimeric promoter 6A8 is functional in human muscle tissue. Luciferase was measured in homogenates of human fetal muscle 2 days after injection of luciferase-encoding plasmids. Results are expressed as mean \pm SEM for 3-6 injections for each clone. ~~Figures~~ **Figure** 8A-8I shows an alignment of the polynucleotide sequences of WT human AD169 and Towne CMV promoters (SEQ ID NOS:19 and 20) and exemplary polynucleotide sequences of the invention (SEQ ID NOS:1-18). The arrow located between the nucleic acid residue positions equivalent to nucleic acid residues 808-809 of the human Towne CMV promoter sequence indicates the transcription start site. The predicted boundary between the first exon and the first intron is also indicated by an arrow between nucleic acid residues 930 and 931 of the human Towne CMV promoter sequence. The last sequence shown in the alignment (SEQ ID NO:21) represents a "consensus sequence" of aligned polynucleotide sequences. The alignment was prepared using the CLUSTALW multiple sequence alignment algorithm, a part of the Vector NTI version 6 sequence analysis software package (Informax, Bethesda, MD). The CLUSTALW program initially performs multiple pairwise comparisons between groups of sequences and then assembles the pairwise alignments into a multiple alignment based on homology. For the initial pairwise alignments, Gap Open and Gap Extension penalties were 10 and 0.1, respectively. For the multiple alignments, Gap Open penalty was 10, and the Gap Extension penalty was 0.05. The protein weight matrix employed was the BLOSUM62 matrix.--

Please replace the paragraph beginning at page 7, line 14, with the following rewritten paragraph:

B2
--~~Figures~~ **Figure** 10A-10D shows an alignment of the polynucleotide sequences of WT of the promoter/enhancer regions of the WT Rhesus monkey (SEQ ID NO: 22), Vervet monkey (SEQ ID NO:23), and human Towne (SEQ ID NO:20) CMV isolates.--

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Please replace the paragraph beginning at page 8, line 18, with the following rewritten paragraph:

B3
--A "chimeric promoter/enhancer" is a non-naturally occurring promoter/enhancer that includes nucleotides from more than one source nucleic acid. The source nucleic acids can be naturally occurring nucleic acids (e.g., nucleic acids from different isolates or species used in family shuffling), but also can be non-naturally occurring nucleic acids. Those of skill in the art will appreciate that the phrase "nucleotides from more than one source nucleic acid" describes the identity of a particular residue at a particular position in a chimeric nucleic acid or the sequence of nucleotides in a particular region of the chimeric nucleic acid. Thus, two polynucleotide sequences in a chimeric nucleic acid are said to be from different source nucleic acids if the polynucleotide sequences are each identical to a polynucleotide sequence in one of the source nucleic acids. This language does not imply that the chimeric nucleic acid was necessarily formed by joining polynucleotide sequences obtained directly from the source nucleic acids, although the invention encompasses chimeric nucleic acids formed in this ~~manner~~ ~~manner~~. As used herein, the term "promoter/enhancer" can refer to either a promoter sequence, as defined above, or an enhancer sequence, or a polynucleotide sequence including both types of sequences. --

Please replace the paragraph beginning at page 32, line 10, with the following rewritten paragraph:

B4
--In one embodiment, variants of SEQ ID NOS:1-18 can be designed based on the properties disclosed herein for these polynucleotides. Thus, for example, the 12C9 polynucleotide sequence (SEQ ID NO:3) lacks CMV promoter nucleic acid residues beyond about nucleotide residue 909, numbered according to the consensus sequence shown in Figures 8A-8I ~~Figure 8~~. Yet this polynucleotide sequence still serves as an efficient promoter of β -galactosidase expression as demonstrated by the in vivo assay for anti- β -galactosidase antibody shown in Figure 6A. This observation indicates that CMV promoter/enhancer sequences downstream (relative to the direction of transcription) of the residue corresponding to residue 909 in the Figures 8A-8I ~~Figure 8~~ consensus sequence are not required for efficient expression of an operably linked transgene. Accordingly, the invention encompasses nucleic acids that include variants of SEQ ID NOS: 1, 2, and 4-18 that lack such downstream CMV promoter/enhancer sequences. In preferred embodiments, such variants include the CAAT box and/or the TATA box (both of these motifs are underlined in

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B4
cont

Figure 8E) present in region corresponding to about nucleotide residues 840-890 of the consensus sequence shown in Figures 8A-8I ~~Figure 8~~. Exemplary nucleic acids of this type lack CMV promoter nucleic acid residues beyond about nucleotide residue 900, 910, 920, 930, and 940, numbered according to this consensus sequence.--

Please replace the paragraph beginning at page 32, line 27, with the following rewritten paragraph:

B5

--The polynucleotide sequences shown in Figures 8A-8I ~~Figure 8~~ include a first exon beginning at about nucleotide residue 810 and extending to about nucleotide residue 932, numbered according to the consensus sequence shown in Figures 8A-8I ~~Figure 8~~. In some application, it may be desirable to delete this sequence. Thus, invention also encompasses nucleic acids that include variants of SEQ ID NOS: 1, 2, and 4-18 lacking these exon sequences. Exemplary nucleic acids of this type lack CMV promoter nucleic acid residues beyond about nucleotide residue 810, 820, 830, 840, 850, 860, 870, 880, and 890, numbered according to this consensus sequence.

Please replace the paragraph beginning at page 39, line 4, with the following rewritten paragraph:

B6

--The chimeric promoter/enhancer polynucleotide sequences, or fragments or variants thereof is joined to nucleic acids that are to be expressed (e.g., coding regions for polypeptides, tRNA and rRNA molecules, antisense nucleic acids, and the like), using techniques that are known to those of skill in the art. Suitable nucleic acids can encode a protein from any organism, e.g., a viral, bacterial, eukaryotic, mammalian, or human protein. Viral proteins of interest include those from dengue virus, human immunodeficiency virus (HIV), Japanese encephalitis virus, Venezuelan encephalitis virus. Examples of nucleic acids that can be incorporated into an expression cassette of the invention include a nucleic acid encoding: an immunogen; an immunomodulatory molecule, such as a co-stimulatory molecule (e.g., B7-1, B7-2, or other polypeptide that binds or associates with a CD28 and/or CTLA-4 receptor); an antigen (e.g., a cancer antigen, such as EpCam/KSA; ~~hepatitis hepatitis~~ B surface antigen or fragment thereof; antigens from hepatitis A, hepatitis C, etc.), including a multivalent or cross-reactive antigen; an adjuvant; an allergen, an antibody; a bacterial toxin, including, e.g., staph/strep enterotoxin and CT/LT (cholera toxin, labile enterotoxin); a cytokine or cytokine receptor (e.g., IL-10 antagonist or receptor); and a prophylactic or therapeutic

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B6
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polypeptide. Other exemplary nucleic acids that can be included in the expression cassettes of the invention include those encoding any of a variety proteins described in commonly assigned PCT Application No. US99/03022 (WO 99/41369), entitled "Genetic Vaccine Vector Engineering," filed February 10, 1999 (106.310WO); commonly assigned PCT Application No. US99/03020 (WO 99/41368), entitled "Optimization of Immunomodulatory Properties of Genetic Vaccines," filed on February 10, 1999 (155.110WO); commonly assigned PCT Application No. US99/03023 (WO 99/41402), entitled "Targeting of Genetic Vaccine Vectors," filed on February 10, 1999 (156.110WO); commonly assigned PCT Application No. US99/02944 (WO 99/41383), entitled "Antigen Library Immunization," filed on February 10, 1999 (157.110WO); commonly assigned PCT Application No. US97/17302 (WO 98/13485), entitled "Methods for Optimization of Gene Therapy by Recursive Sequence Shuffling and Selection," filed September 26, 1997 (107.410WO); commonly assigned PCT Application No. US00/16984 (WO 00/00234), entitled "Methods and Compositions for Engineering of Attenuated Vaccines," filed June 20, 2000 (133.110WO); each of which is incorporated herein by reference in its entirety for all purposes.--

Please replace the paragraph beginning at page 41, line 11, with the following rewritten paragraph:

B7

--In one embodiment, a population of cells comprising a nucleic acid of the invention operably linked to a transgene encoding a polypeptide is used for recombinant protein production. Thus, the chimeric promoter/enhancers of the invention or fragments or variants thereof can be used to express a transgene in any application ~~any application~~ in which expression of the encoded polypeptide is desired. Examples include research applications, e.g., where the polypeptide is expressed in functional studies; any application, including in vitro or in vivo research or diagnostic ~~diagnostic~~ assays, in which expression of a marker polypeptide is desired. In vivo applications, including gene therapy and genetic vaccination are discussed in greater detail below. The nucleic acids of the invention can also be used to produce any polypeptide of interest for research, medical, or industrial use.--

Please replace the paragraph beginning at page 46, line 3, with the following rewritten paragraph:

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B8
--The recombinant optimized promoters of the invention can also be used in conjunction with optimized antigens. Types of wild-type antigens that can be employed for various conditions and for use in genetic vaccines are described in commonly assigned PCT Application No. PCT/US99/02944 (WO 99/41383), entitled "Antigen Library Immunization," which is incorporated herein by reference in its entirety for all purposes. Furthermore, multiple antigens can be expressed from a monocistronic or multicistronic form of the vector comprising at least one recombinant promoter of the invention. ~~Moreover~~ ~~Moreover~~, an antigen for a particular condition can be optimized using recombination and selection methods analogous to those described herein. Such methods, and antigens appropriate for various conditions, are described in PCT Application No. PCT/US99/02944.--

Please replace the paragraph beginning at page 48, line 9, with the following rewritten paragraph:

B9
--In vivo indirect contact/administration formats, the nucleic acid or vector is typically administered or transferred indirectly to the cells to be treated or to the tissue site of interest, including those described above (such as, e.g., skin cells, organ systems, lymphatic system, or blood cell system, etc.), by contacting or administering the nucleic acid or vector of the invention directly to one or more cells or population of cells from which treatment can be facilitated. For example, tumor cells within the body of the subject can be treated by contacting cells of the blood or lymphatic system, skin, or an organ with a sufficient amount of the polypeptide such that delivery of the nucleic ~~nucleic~~ acid or vector to the site of interest (e.g., tissue, organ, or cells of interest or blood or lymphatic system within the body) occurs and effective prophylactic or therapeutic treatment results. Such contact, administration, or transfer is typically made by using one or more of the routes or modes of administration described above.--

Please replace the paragraph beginning at page 56, line 27, with the following rewritten paragraph:

B10
--Four strains of cytomegalovirus (CMV) were obtained from American Type Culture Collection (ATCC) (Rockville, MD). Human AD169 (VR-538; Rowe W. et al. (1956) *Proc. Soc. Exp. Biol. Med.* 92:418 445:794-801) and Human Towne (VR-977; Plotkin SA (1975) *Infect. Immun.* 12:521-27) strains were isolated from human patients with CMV infections, while the 68-1

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cont

(Asher DM (1969) *Bacteriol. Proc.* 269:91) and CSG (Black H (1963) *Proc. Soc. Exp. Biol. Med.* 112:601) strains were isolated from Rhesus and Vervet monkeys, respectively.--

Please replace the paragraph beginning at page 58, line 18, with the following rewritten paragraph:

B11

--For a description of the human CMV (hCMV) promoters, *see, e.g.*, US Pat. No. 5,385,839 and Meier, J., *et al.*, *Intervirology* 39:331-342 (1996), the full disclosure of which is incorporated herein by reference in its entirety for all purposes. For cloning procedure for a hCMV and Rhesus CMV promoter, *see, e.g.*, US Pat. No. 5,385,839 and Alcendor *et al.*, *Virology* 194:815-812 (1993), the full disclosure of each of which is incorporated herein by reference in its entirety for all purposes. The nucleotide sequences for human CMV promoters, Towne and AD169 strains, are shown in Figures 8A-8I ~~Figure 8~~. The sequence for human CMV promoter Towne strain is shown at GenBank Accession No. X03922. The nucleotide sequences for the Rhesus and Vervet monkey CMV promoters are shown in Figures 10A-10E ~~Figure 10~~. Rhesus CMV IE promoter is shown in Alcendor *et al.*, *Virology* 194:815-812(1993). AGM CMV IE (Colburn strain) is shown at GenBank Accession No. M16019.--

Please replace the paragraph beginning at page 69, line 19, with the following rewritten paragraph:

B12

--Mice were injected with 10 µg β-galactosidase-encoding plasmids on days 0 and 15, and serum collected on days 14 and 28 for measurement of anti-β-galactosidase antibodies. Plasmids comprising a AD169, Towne, or Vervet parental nucleic acid sequence operably linked to β-galactosidase nucleic acid sequence were also injected into groups of mice in a similar manner and used for comparison with the plasmids comprising the chimeric promoter sequences. As a vector control, an empty vector comprising a promoterless β-galactosidase-encoding plasmid (pcDNAβ-gal) was injected into mice in a similar manner. A group of mice that were not inoculated with any vector served as a control group. Figures Figure 6A and 6B shows the antibody titer levels measured in serum by ELISA methods, where the serum was obtained following injection of mice with β-galactosidase-encoding plasmids (10 µg or 4 µg plasmid, respectively) at the time (day) noted above.--

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Please replace the paragraph beginning at page 70, line 23, with the following rewritten paragraph:

B13

--Sequence analysis of selected shuffled chimeric promoters revealed that they comprised mainly nucleic acid sequences from the AD169 and Towne human parental nucleic acid sequences. In addition, the sequences contain between 2 and 17 unique nucleotides throughout the promoter. Deletions of one or two nucleotides occur in several of the clones, and 11E2 also has an additional nucleotide (nt) after nt853 (numbering is based on the consensus sequence as shown in Figures 8A-8I Figure 8). Clones 6F6, 9G7, 11E2, and 12C9 contain nucleotide sequences derived from the Rhesus monkey exon A approximately from nt817 (which is close to the transcription start site) to nt863. Clones 4B5, 6B2, 6D4, and 12E1 have a deletion corresponding to the region 684-735 nucleotides in the consensus sequence. Clone 12C9 is truncated at nucleotide (nt) residue 909 (numbered according to the consensus sequence shown in Figures 8A-8I Figure 8). Notably, clone 12C9 gave a comparable or increased antibody response in the B-gal screening assay relative to other chimeric clones or the parental sequences despite having a truncated sequence. Compared with the human AD169 and Towne nucleic acid sequences, the 12C9 nucleic acid sequence lacks a short segment of the nucleic acid sequence corresponding to the first exon and intron of each of the AD169 and Towne strains.--

Please replace the paragraph beginning at page 71, line 9, with the following rewritten paragraph:

B14

--There is also a deletion in clone 9E1 corresponding to nucleotides 319 to 512 in the parental clones. In all of the shuffled sequences, the TATA box (or TATATAA box), CAAT (or CAAAT box) box and transcription start site (T=thymine, C=cytosine, A=adenine nucleotide bases) are identical to those found in the AD169 and Towne parental sequences (see Figures 8A-8I Figure 8). For known CMV promoters, it is generally believed the TATA box is important for promoter activity.--